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ANATOMICAL AND OTHER STUDIES ON MAZZARD CHERRY SEEDLINGS HAVING EXCESSIVE ROOTS AT THE COLLAR REGION [1]

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INTRODUCTION

Malformations characterized by an excessive number of roots at the collar region of cherry trees were observed in a nursery in 1929. The condition was noted on approximately 3 percent of the trees that had been budded on mazzard cherry (*Prunus avium* L.) seedlings. These tufts of roots (fig. 1, *A*) on the seedling part of the tree presented such an abnormal and unsightly appearance that the grower discarded the trees. Although these malformations bore a superficial resemblance to the hairy root disease caused by *Phytomonas rhizogenes* Riker et al., common on apple (*9*),[3] they were not typical of this disease, and routine pathological studies failed to disclose a causal organism.

Several years later, excessive roots just below the ground level were observed on a small percentage of 1-year-old mazzard seedlings grown at Beltsville, Md. These tufts of roots (fig. 1, *B* and *C*), in contrast with normal rooting (fig. 1, *D*), were obviously an early expression of the condition that had been observed previously on the nursery trees. As the presence of numerous root primordia in the young hypocotyl obviously would account for the excessive-root malformations observed on the trees in the nursery, anatomical studies were made on young mazzard cherry seedlings. Such studies are of considerable importance in supplying information for interpreting growth responses in plants (*3*).

Among horticultural workers, Priestly and Swingle (*8*), in particular, have emphasized the value of and the need for anatomical studies as a basis for explaining the vegetative responses of certain plants propagated by root or stem cuttings. Their detailed studies and their extensive review of the literature on the role of adventive root and shoot primordia in vegetative propagation provide an excellent background for workers interested in studying the ontogeny of these organs. Information of practical value is accumulating as a result of anatomical analyses that have served to explain such responses as the rooting potentialities of etiolated shoots (*7*), regeneration in stem and root cuttings (*2, 5, 6, 13*), and the dwarfing effect of certain rootstocks (*1*). On apple, anatomical studies have been made on the origin of root and shoot primordia in stem tissue (*5, 14*) and in roots, particularly on regeneration (*12*) and the initiation of primordia (*10*).

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[2] The writer is indebted to J. J. Bowman, of this Division, for assistance in preparing the slides and photomicrographs for these studies.

[3] Italic numbers in parentheses refer to Literature Cited, p. 15.

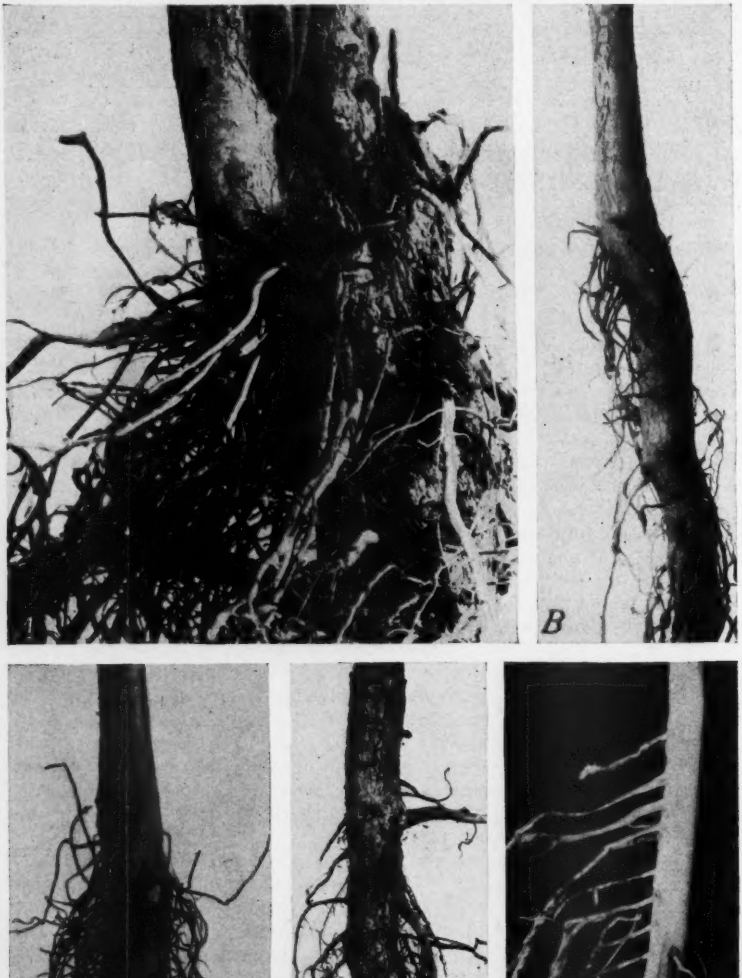

FIGURE 1.—Root development on the collar region of mazzard cherry: *A*, Tufts of roots on a 1-year-old budded tree; *B* and *C*, excessive roots on 1-year-old seedlings; *D*, root development on a normal 1-year-old seedling; *E*, linear arrangement of primary roots on a 6-week-old seedling (slightly enlarged).

The anatomical studies reported herein were intended mainly to supplement the present information on the origin and development of root primordia in fruit trees (*4*). An effort was made to identify early stages of root primordia and to note any synchronization of the initiation of primordia with the ontogenetical development of the parent tissue (*4*, *8*). The limited pathological studies reported were made mainly to eliminate the suspicion that the hairy root organism may be a causal factor in the development of these excessive roots.

MATERIAL AND METHODS

In the pathological studies, routine methods in isolation and in inoculation tests were used. Supplemental observations were also made on seedlings grown in steamed soil.

For the anatomical studies, mazzard seeds were germinated and the seedlings were grown in the greenhouse in order to observe the occurrence of excessive roots. When the seedlings had made shoot growth ranging from 8 to 15 cm., portions of the hypocotyl and root were killed in formalin-acetic-acid-alcohol solution. The material was later embedded in paraffin, sectioned, and stained in the usual histological routine, except that butyl alcohol was frequently used in the killing as well as in the dehydrating series.

OBSERVATIONS AND RESULTS

PATHOLOGICAL EXPERIMENTS

The results of the limited pathological experiments and observations are summarized briefly. The malformations (fig. 1, *A*) not only are atypical of the hairy root disease, but also they occur in districts where the hairy root organism is not commonly found. The general aspect of these excessive root formations more closely resembles the genetic condition known as noninfectious hairy root on apple (*11*), but there is no evidence that the condition on cherry is genetic.

Attempts to isolate the hairy root organism from affected trees over a 5-year period were unsuccessful except in one instance, which was considered fortuitous because of previous experience with this organism as a surface contaminant (*9*). Inoculations with the hairy root organism into the collar regions of 2-month-old mazzard cherry seedlings growing in previously steamed soil, however, resulted in infection on 13 of the 15 seedlings that were inoculated; the 10 punctured, noninoculated seedlings remained normal. The symptoms on these infected seedlings were typical of the early stages of hairy root on apple and were characterized by dense aggregations of root "knobs"; the organism was reisolated. Although this evidence establishes mazzard cherry as a host for the hairy root organism, it also indicates that the type of malformation under consideration in this paper is not the result of infection by this organism. Because the possibility that the condition is the result of infection by a bacterium or a fungus appeared so remote, subsequent attention was confined to morphological and anatomical features as a supplement to previous studies on the origin of adventive primordia (*10*).

MORPHOLOGICAL AND ANATOMICAL FEATURES

Vascular Arrangement

The hypocotyl of mazzard cherry seedlings seldom exceeds 7 cm. in length; under conditions favorable for extension, the maximum observed length was 9.5 cm. Under greenhouse conditions favorable for vigorous growth, some of the young seedlings exhibited a longitudinal splitting in the cortex of the hypocotyl (fig. 1, *E*). Lateral roots also frequently made their appearance normally in vertical rows on the hypocotyl. This linear arrangement of course indicates a morphological relation of these roots to the xylem in the hypocotyl (*3*, *14*). As expected, adventive root primordia were found in such seedlings in sufficient numbers to account for the masses of roots that sometimes occur on older nursery trees (fig. 1, *A*) and on 1-year-old seedlings lined out for budding (fig. 1, *B* and *C*).

The illustrations in figure 2 are transverse sections of a seedling showing the vascular arrangement at different zones in the root and hypocotyl of a seedling beginning secondary growth. The relative size of the vascular cylinder conforms with the gross appearance of seedlings, which at this stage generally are considerably enlarged at the base of the hypocotyl.

In the root (fig. 2, *A*), the xylem cylinder is established and the epidermal cells are collapsing, preliminary to the disintegration of the cortex.

At the base of the hypocotyl (fig. 2, *B*), the xylem cylinder is interrupted by four gaps, resulting in four arcs of xylem. Successive sections up the hypocotyl show a progressive tendency toward forking into two separate strands at the center of each primary xylem group. At the zone shown, tangential divisions in the layer of cells of the pericycle inside the endodermis indicate the initiation of the phellogen. The zone 7 mm. farther up the hypocotyl (fig. 2, *C*) is characterized by a definite gap in the center of each arc of xylem. At this level gaps in the procambium strands from the stem apical meristem occur over the four primary xylem regions. Procambium activity between the four "paired" xylem strands results in xylem differentiation, which serves to unite one strand from each of the "paired" xylem strands with the adjacent strand of the next pair. This general type of orientation in the transition zone corresponds with "type A" of Eames and MacDaniels (*3*). At this stage the outline of the xylem is roughly in the form of a square. The four-sided appearance is accentuated by the presence of thick-walled parenchyma in tangential lines over the cambium. These thick-walled parenchyma cells possibly serve as protective tissue previous to the differentiation of four arcs of fibers, which at a slightly later stage (fig. 3) form a cap over the phloem elements.

Figure 2.—Transection showing vascular arrangement at different levels in a young seedling beginning secondary growth: *A*, Root tissue just below base of hypocotyl; *B*, tissue at the base of the hypocotyl, 1.1 mm. above *A*; *C*, tissue in a zone 7 mm. farther up the hypocotyl. All × 80. *e*, epidermis; *en*, endodermis; *p*, pericycle; *par*, thick-walled parenchyma; *r*, lateral root primordium; xy_1, region of primary xylem.

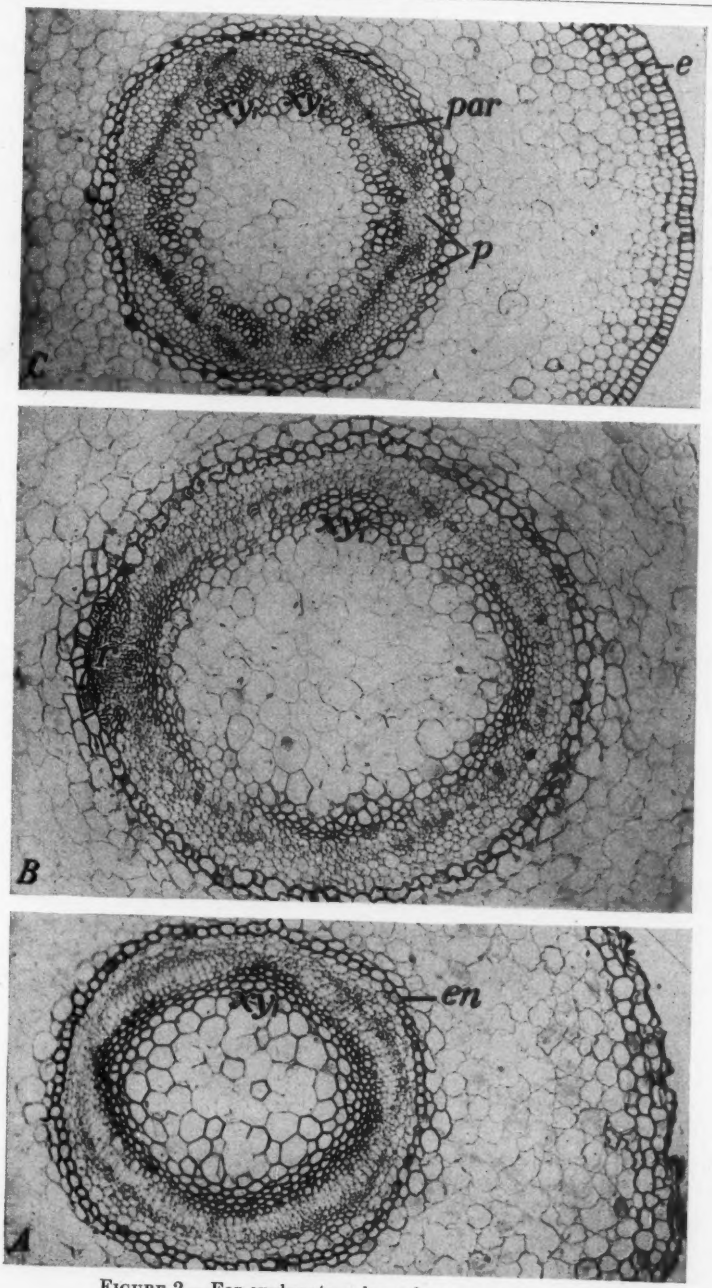

FIGURE 2.—For explanatory legend see opposite page.

Lateral Roots

The origin and development of lateral roots are described because some such roots are undoubtedly intermingled with adventive roots in the tufts of roots under discussion.

Lateral roots develop in the pericycle over the general region of the primary xylem at the root-stem junction (fig. 2, *B*) and in the hypocotyl (fig. 3). As a result, these roots appear in vertical rows. The primordia shown in figure 4, *A* and *B*, are developing at the base of the hypocotyl. At this comparatively advanced stage many cells in this meristematic area have large deep-stained nuclei. Activity of this type occurs over a comparatively large area (fig. 4, *A*) even including the enveloping endodermis. The meristematic endodermis persists as an enveloping layer in at least considerably later stages than that shown in figure 4, *B*.

When conditions of growth are such that the individual strands in each of the "paired" xylem strands are widely separated, lateral roots

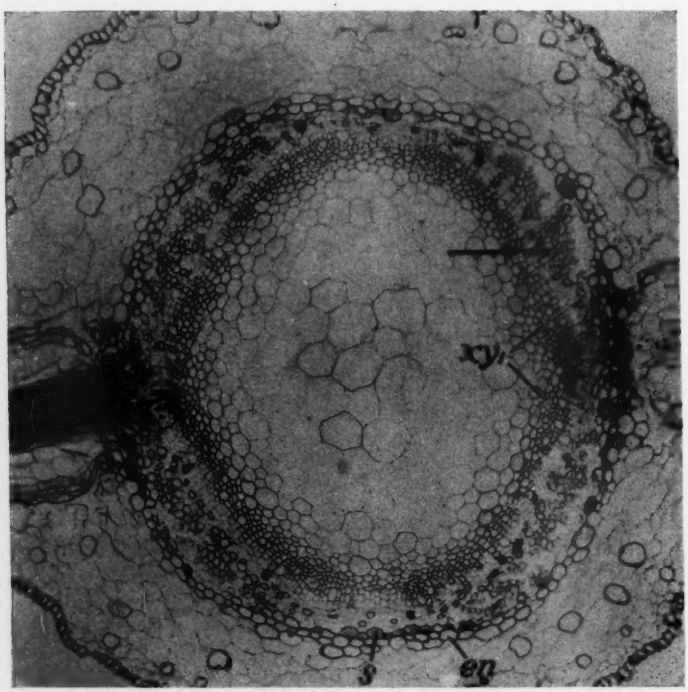

Figure 3.—Transection of a hypocotyl at a zone about 3.5 cm. above the base, at an early stage in secondary growth. The relation of the two lateral roots to the "paired" primary xylem strands (xy_1) is shown. An adventive primordium (indicated by arrow) lies beneath one of the four groups of fibers (*f*) (see fig. 8, *A*). Slight suberization in the layer of pericycle cells beneath the endodermis (*en*) is indicated at *s*. × 80.

may originate in pericycle tissue in a median position between the two protoxylem regions (fig. 4, *C*). The point of origin of this lateral root with reference to the xylem strands, which are alined tangentially at this level, indicates that the inherent potentiality of the pericycle cells over and between these protoxylem regions is an important factor in determining the initiation of primordia.

Adventive Root Primordia

At an early stage in secondary growth of the root, near the base of the hypocotyl adventive primordia are formed from a small group of cells in the cambium-phloem region slightly to one side of the protoxylem (fig. 5). No effort was made to determine whether or not pri-

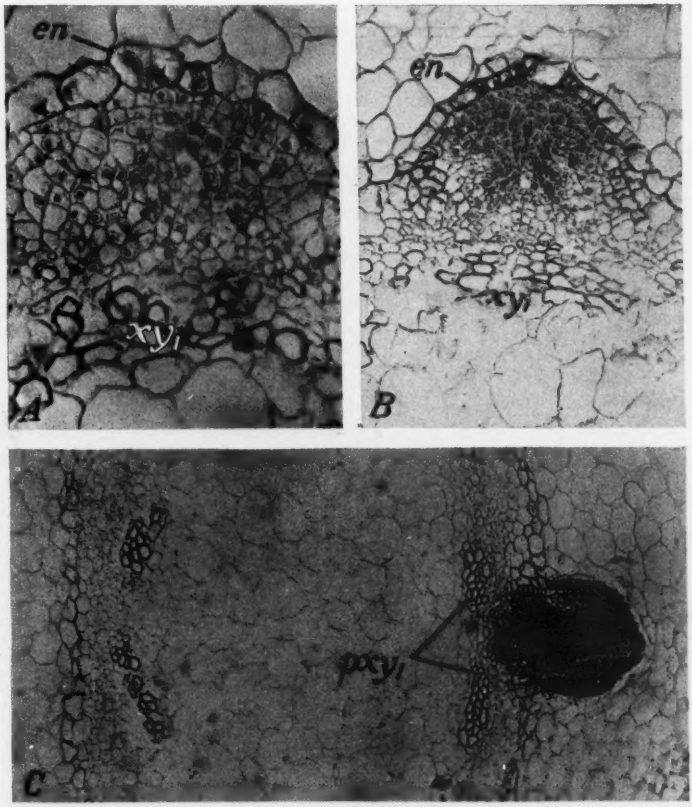

FIGURE 4.—*A* and *B*, Transections of lateral-root primordia at a zone near the base of the hypocotyl: *A*, Stage in which cell activity is evident over a comparatively large area, × 350; *B*, later stage characterized by rapid cell division with comparatively little cell enlargement, × 175. *en*, endodermis; xy_1, primary xylem region. *C*, Transection at the base of a hypocotyl showing position of origin of a lateral root with reference to the protoxylem regions (pxy_1) of the tangentially arranged primary xylem strands, × 90.

mordia developed in the root in other regions at either this or later stages.

In the hypocotyl adventive primordia are initiated in early stages of secondary growth and continue to be formed, with less frequency, however, in relatively late stages of secondary growth. At the early stages of secondary growth these primordia were observed (1) in the general region over the "paired" strands of primary xylem and (2) in the cambium-secondary-phloem region beneath the groups of fibers.

Progressive stages in the development of the primordia that originate in the general region over the "paired" primary xylem strands are shown in transverse sections in figure 6, *A–E*, and in longitudinal sections in figure 7, *A–D*. This region is characterized by a narrow zone of thin-walled meristematic cells that are presumably nondifferentiated derivatives of the pericycle. A cambium frequently develops in this region, but it does so comparatively late and after a considerable amount of xylem and phloem elements have been differentiated in each of the quadrants between the four "paired" primary xylem strands. Adventive primordia are initiated soon after the beginning of cambium activity in this region. The initiating cells are thin-walled and contain prominent nuclei, and most of them appear to be recent cambium derivatives. Growth and further development of the primordium up to the stage shown in figure 6, *E*, are largely the result of additions from the cambium, as can be observed in figure 6, *C* and *D*.

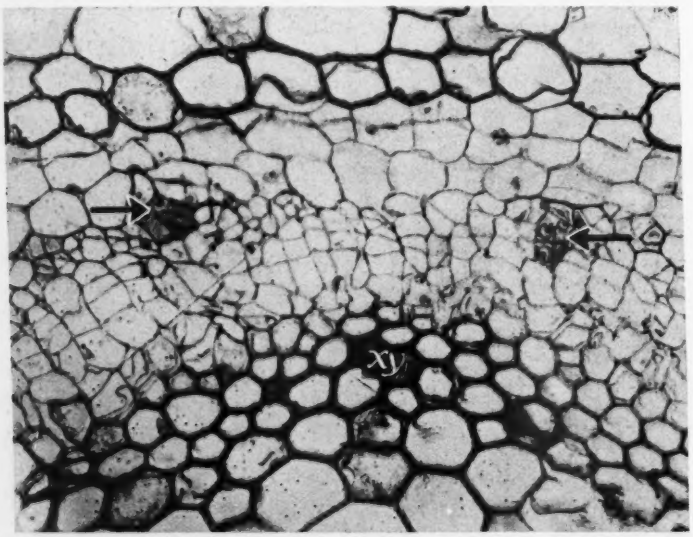

FIGURE 5.—Transection of a root immediately below the hypocotyl, showing early stages of adventive primordia (indicated by arrows) and their position relative to the primary xylem (xy_1), × 450.

A protective covering over the meristem is formed as a result of divisions in the phellogen, which has developed in the pericycle layer of cells beneath the endodermis (figs. 6, *A–D*; 7, *C* and *D*). In the areas over the young primordia the cell walls of these pericycle deriva-

tives are thick and heavily stained. In this connection it is of interest to note that there is a tendency toward suberization in the outer layer of the pericycle over the primary xylem areas (fig. 3) at an early stage when the pericycle over the four arcs of fibers is still active in differentiating the pericycle fibers.

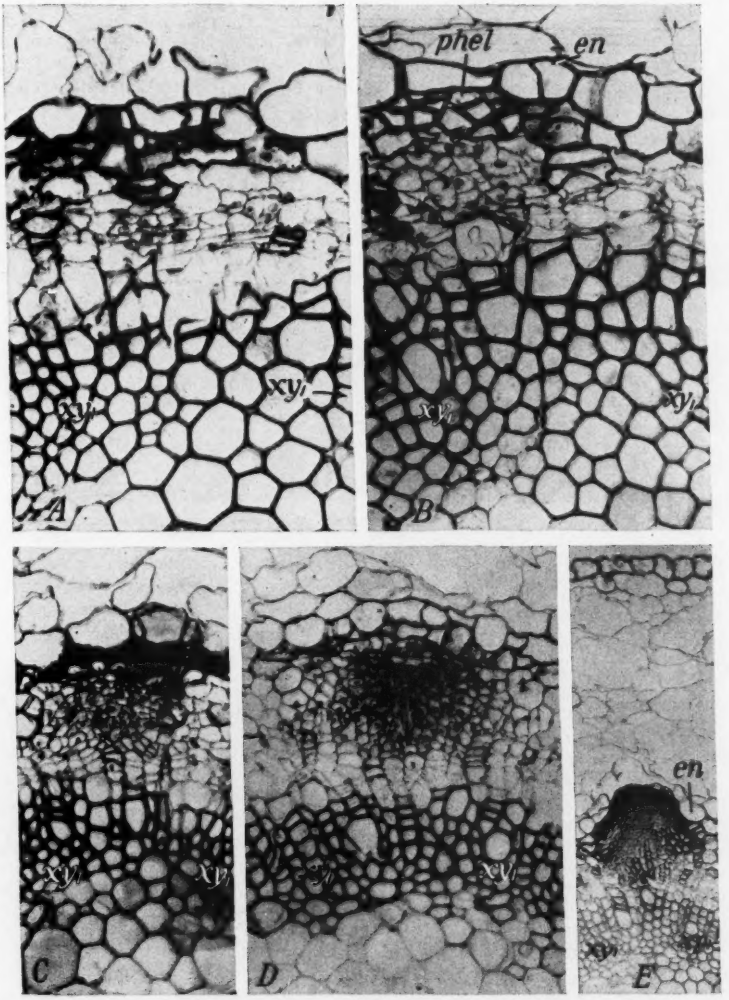

FIGURE 6.—Transections of hypocotyls showing the position of adventive primordia in the general region over the "paired" primary xylem strands and progressive stages in the development of primordia that have been initiated during early stages of secondary growth. xy_1, region of primary xylem; *en*, endodermis; *phel*, phellogen activity. *A* and *B*, × 400; *C* and *D*, × 225; *E*, × 110.

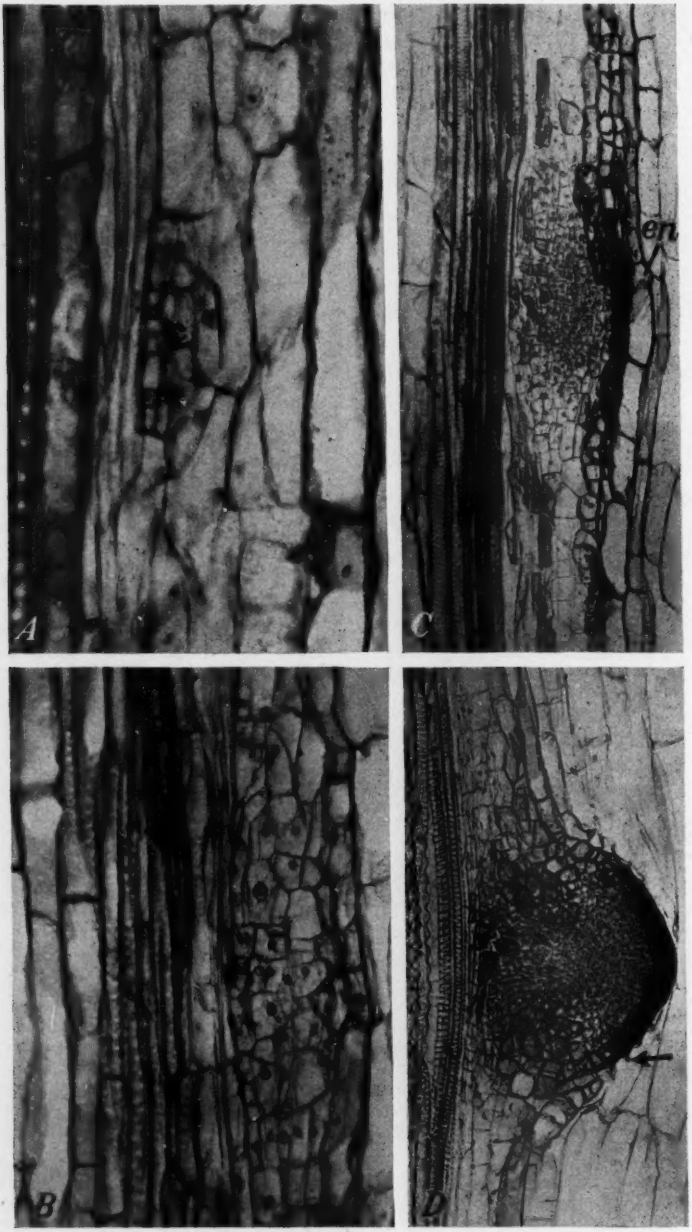

FIGURE 7.—For explanatory legend, see opposite page.

At the beginning of secondary growth, primordia are also initiated under these groups of fibers, as shown (by arrow) in figure 8, *A*, which is part of figure 3 enlarged, and in figure 8, *B*. A close series showing the progressive development of these primordia is shown in figure 8, *C–G*. Although the position of the large nucleated cell and the presence of the two nuclei in the cambium region beneath it suggest that the single large nucleated cell shown in figure 8, *C*, marks the initial stage of a primordium, positive identification is probably not warranted. In general, it is evident that most of these primordia are initiated in the cambium region from direct cambium derivatives. In only rare instances were there indications that the primordia originated from partially matured cells in the phloem (fig. 8, *D*). That the primordium may actually be "deep-seated" with respect to the present line of the cambium, because of intense meristematic activity with but little cell-wall delineation, is shown in figure 8, *B*.

With increasing vascular differentiation, activity becomes pronounced in the phellogen over the entire periphery. Relatively few primordia are formed after secondary growth has progressed much beyond the stage shown in figure 9, *A*, in which periderm is being formed. Those primordia that develop comparatively late originate in or near the phloem ray parenchyma and are usually initiated as a result of activity in a comparatively large group of cells (fig. 9, *A*, *B*, and *C*, *b*). Such cells lie in the general radial planes that pass through the primary xylem regions, in which planes lateral (fig. 9, *C*, *a*) and adventive roots (fig. 9, *C*, *c*) may have previously developed.

DISCUSSION AND CONCLUSIONS

The type of malformation discussed herein differs from that of infectious hairy root on apple and other hosts, in that these root formations occur over a general and indefinite region as compared with the more compact and localized masses of root primordia or of roots resulting from infection by the hairy root organism (*9*). Further study would be required to determine whether genetic or environmental factors are involved; limited observations indicate, however, that conditions favoring vigorous growth frequently may result in numerous roots on the hypocotyl of young seedlings. In the absence of information to the contrary, it is assumed that the economic losses due to discarding affected trees are relatively minor. The evidence, however, that the malformations described herein are nonpathogenic should aid nurserymen and inspectors in grading cherry trees.

In the anatomical studies an attempt was made to show the plan or pattern along which the hypocotyl develops as a result of activity of specialized tissue. Thus, four pairs of procambium strands in the upper levels of the transition zone become broadened tangentially in the older tissue at lower levels. At the base of the hypocotyl and in the adjacent root tissue the procambium forms an almost continuous sheath. The differentiation of the procambium into primary xylem

FIGURE 7.—Radial sections of hypocotyls in vertical planes passing through the general region of the "paired" primary xylem strands, showing origin and development of adventive primordia. Phellogen activity is evident beneath the endodermis (*en*), and suberization of the cells forming a protective covering is pronounced. The arrow in *D* indicates remnants of cell walls of the collapsed endodermis. *A*, × 650; *B*, × 350; *C*, × 250; *D*, × 150.

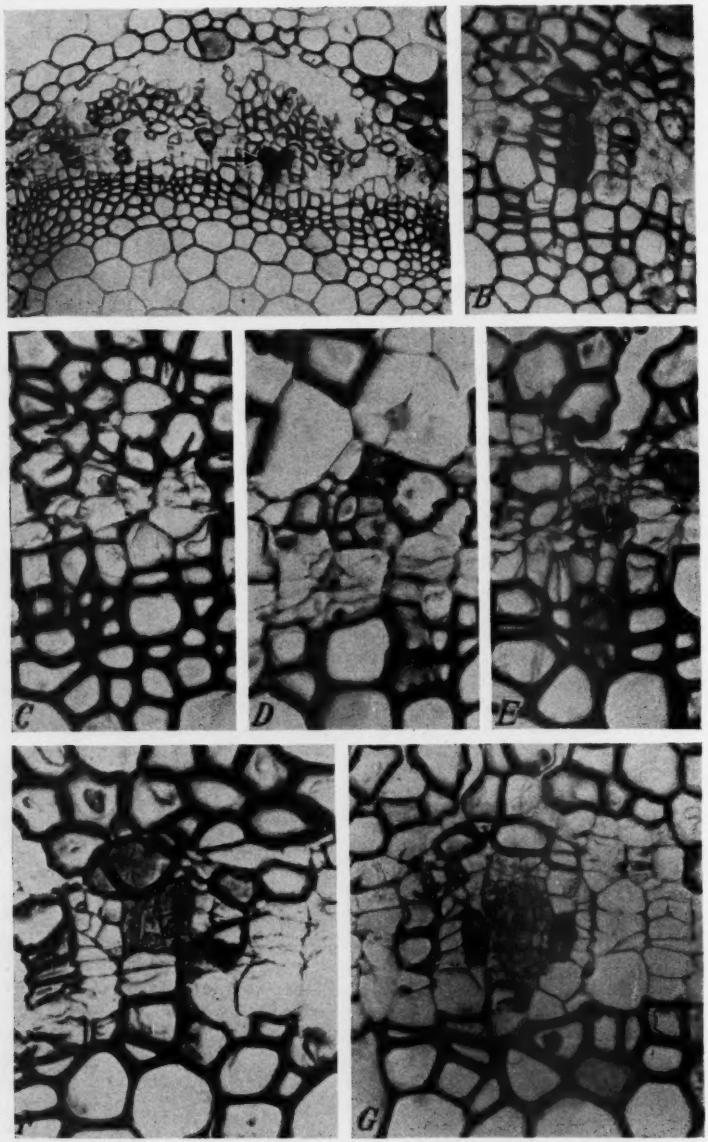

FIGURE 8.—Transections of hypocotyls showing (*A* and *B*) the adventive primordia that are initiated beneath the fibers (*f*) and (*C*–*G*) progressive stages in the development of such primordia. *A*, × 125; *B*, × 350; *C*–*G*, × 650.

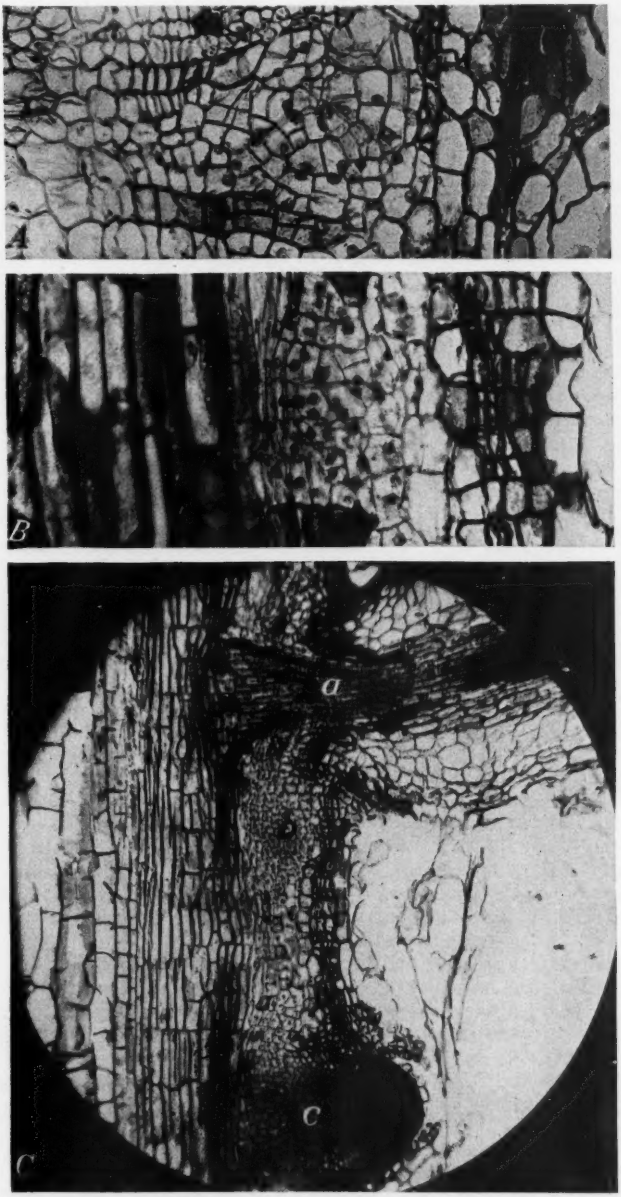

FIGURE 9.—Sections of hypocotyls showing origin of adventive primordia in relatively late stages of secondary growth: *A*, Transection, × 350; *B*, radial section, × 350; *C*, radial section, × 50. *a*, lateral root; *b* and *c*, adventive primordia.

elements therefore results in four broad arcs of xylem near the base of the hypocotyl, while nearer the upper level of the transition zone definite radial gaps are present in each pair of primary xylem strands. These "paired" primary xylem strands sometimes appear as widely separated entities (fig. 4, *C*), but more frequently the metaxylem elements of each of the "paired" strands tend to form a tangential line with the metaxylem elements of one strand from the adjacent "paired" strands. No attempt was made to determine an average for the length of the transition zone.

The vertical radial plane passing through the central regions of the broad primary xylem strands in the roots passes through the central region between each pair of primary xylem strands in the hypocotyl. Lateral root primordia are developed in the pericycle tissue in this plane. This area between and over the protoxylem strands is occupied by a meristem of pericycle which persists for a relatively long time; in other areas at the same level cambium and fibers have been differentiated. It can be reasoned that the inherent potentialities of these cells in which root primordia have their genesis are at least as important as the position of these cells in the organism.

At an early stage in secondary growth (fig. 3) the pericycle fibers are rapidly maturing. The bulge in the endodermis over each group of fibers presumably is the result of activity of pericycle cells in these regions; the pericycle cells over the four "paired" primary xylem strands remain as undifferentiated and comparatively inactive meristematic tissue. At this stage, no indication of a phellogen appears, but at a lower level, immediately below the root-stem junction, a phellogen is established by tangential divisions in the layer of cells of the pericycle underlying the endodermis very early in secondary growth. As is commonly observed, this makes for relatively early periderm formation and disintegration of the cortex in the root.

At the beginning of secondary growth adventive primordia are developed in meristematic tissue from cells that, like the pericycle cells over the primary xylem region, are relatively the least stabilized in differentiating other specialized tissue. Those primordia developing beneath the fibers are initiated in a cambium region from which relatively few xylem elements have been cut off; those primordia that develop in the strips of pericycle over the "paired" primary xylem regions are also initiated near or in a cambium region which has but recently been formed and in which ray parenchyma is common.

Relatively few primordia were formed in later stages of secondary growth after the periderm had become definitely established (fig. 9, *C*). These primordia were confined to a region in the broad phloem rays opposite the primary xylem region—a location in which numerous lateral and adventive roots occur earlier in the ontogeny of the hypocotyl. Relatively large numbers of partially matured cells appear to be involved in the initial stages of these primordia. These cells, however, are genetically related to a cambium, which is differentiating ray parenchyma only.

In almost all cases observed, the outermost cells of all the primordia abut on cells with thickened walls. Possibly such cells function as a mechanical barrier that prevents the loss of essential materials as has been suggested for the role of the endodermis (*7*); they may also serve to initiate meristematic activity as a result of pressure on the underlying meristematic cells. The slight tendency for suberization of

pericycle cells over the primary xylem regions indicates at least that cell-wall thickening precedes the initiation of primorida, but it is recognized that pressure exerted by the growing meristem of the primordia might also produce the same effect.

SUMMARY

Pathological and anatomical studies were made to determine the nature of malformations characterized by an excessive number of roots on the mazzard cherry seedling rootstocks just below the ground line. In extreme cases these tufts of roots cause malformations so pronounced as to result in the discarding of the affected trees.

Although these root formations bear a slight resemblance to the hairy root disease of apple, caused by *Phytomonas rhizogenes* Riker et al., routine pathological studies have failed to disclose a causal organism. Inoculations on young mazzard seedlings with the hairy root organism resulted in infections that were typical of the hairy root disease but atypical of the root formations herein reported.

The anatomical studies demonstrated the presence of root primordia in the hypocotyl of young seedlings in sufficient numbers to account for the gross appearance, observed later, on nursery trees. The fact that many of these primordia bear such a definite morphological relationship to the parent tissue indicates that their presence is a normal occurrence in many seedlings.

Adventive primordia are formed in the hypocotyl most frequently at the beginning of or in the early stages of secondary growth. At this time they become differentiated (1) opposite the general regions of the primary xylem strands as a result of activity in a group of cells most of which appear to be cambium derivatives and (2) in the cambium region beneath the fibers as a result of activity of a small group of cells that are very recent cambium derivatives.

In later stages of secondary growth, adventive primordia are usually confined to the broad phloem rays opposite the primary xylem regions. They are differentiated as a result of activity of a comparatively large number of parenchyma cells.

The morphogenetic relation of the initiating cell or group of cells as well as the environment in which these cells develop has been briefly considered. This relation may be a factor in influencing the production of primordia. In general, adventive primordia are initiated in derivatives from those regions of the cambium that may be considered the least stabilized in producing xylem and phloem elements.

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EFFECTIVENESS AGAINST THE CALIFORNIA RED SCALE OF CUBE RESINS AND NICOTINE IN PETROLEUM SPRAY OIL [1]

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INTRODUCTION

The increasing difficulty of controlling the California red scale (*Aonidiella aurantii* (Mask.)) with hydrocyanic acid gas in parts of the citrus-producing area of California has brought about a serious problem in insect control (*9*).[2] Although oil emulsions are widely used in spraying citrus trees, it is difficult to secure a satisfactory kill of the scales on older wood with concentrations that are safe for the trees (*4*, *5*), particularly in heavy infestations.

In an effort to improve control practices, materials that might be expected to confer added toxicity to the sprays have been incorporated with the oils. Combinations of this kind have been employed in combating a number of other insects. Nicotine has been extensively used with oil.[3] Kagy and Richardson (*7*) showed that solutions of dinitro-o-cyclohexylphenol in oil were toxic to the San Jose scale (*Aspidiotus perniciosus* Comst.). L. H. Dawsey and the writer,[4] in work with mealybugs and the Florida red scale (*Chrysomphalus aonidum* (L.)) at New Orleans, La., found that the effectiveness of oil sprays was enhanced by the addition of rotenone, nicotine, or pyrethrum. In further work at Wooster, Ohio, with the Mexican mealybug (*Phenacoccus gossypii* Towns. and Ckll.) and the willow scurfy scale (*Chionaspis salicis-nigrae* Walsh), they showed that the addition of nicotine and rotenone increased the effectiveness of oil sprays. Although Smith (*10*) was unsuccessful in attempts to increase the toxicity of spray oils to the California red scale by the addition of various organic compounds, La Due (*8*) has reported that the addition of derris resins to oil with intermediary solvents gives increased toxicity to several scale insects of citrus.

The experiments reported herein include field tests of nicotine and cube resins with petroleum oil applied to the California red scale on lemon trees. The primary objectives were to determine whether the effectiveness of oil sprays could be improved by the addition of these two toxicants and, having demonstrated that point, to compare their relative effectiveness at selected concentrations.

[1] Received for publication June 5, 1942.

[2] Italic numbers in parentheses refer to Literature Cited, p. 25.

[3] MCINDOO, N. E., ROARK, R. C., and BUSBEY, R. L. A BIBLIOGRAPHY OF NICOTINE. SECT. 2. THE INSECTICIDAL USES OF NICOTINE AND TOBACCO. U. S. Bur. Ent. and Plant Quar. Cir. E-392, 358 pp. 1936. [Processed.]

[4] Unpublished.

MATERIALS AND METHODS

All sprays contained a heavy mineral oil having the following specifications:

Saybolt viscosity at 100° F.	85 seconds.
Unsulfonatable residue	94.6 percent.
Specific gravity	0.858.
Percent distilling at 636° F.	5 percent.
Volatility (24 hours at 100° F.)	1.2 percent.

The cube resins contained 22.3 percent of rotenone and were completely soluble in carbon tetrachloride. Because of their limited solubility in petroleum oil, they were first dissolved in an intermediate solvent consisting of 1 part (by volume) of trichloroethylene and 2 parts of dibutyl phthalate. Ten parts of this solution were added to 90 parts of petroleum oil before emulsification, after which the resins were present partly in solution and partly in suspension in the oil.

Nicotine from a lot containing 95 percent of free base was used for the sprays containing this toxicant and was added to the spray mixture at the time of filling the tank.

To make the oil phase identical in all tests, the trichloroethylene-dibutyl phthalate mixture was always included in the oil phase, and all oil concentrations mentioned hereafter include 10 percent of this intermediate solvent. For example, a spray, with or without toxicant, that is said to contain 1 percent of oil included 0.9 percent of petroleum oil and 0.1 percent of the solvent mixture. Stock emulsions were prepared the day before spraying by agitating equal volumes of oil and an aqueous solution of ground bone glue with a motor stirrer for 20 minutes.

Spray applications were made with a small power sprayer, operated at 300 pounds' pressure, with a spray gun having a disk opening 3/32 inch in diameter. Trees were first sprayed from the inside and then circled on the outside from the ground. Tops of the trees were sprayed from a tower.

Oil deposits were determined according to the method described by Dawsey and Hiley (*3*) with the modification that the final drying was made at 55° C. while a slow stream of air was forced into the flasks. Each sample consisted of 80 or 100 disks 1.5 cm. in diameter, 4 from each of 20 or 25 leaves from a single tree. One or more samples were taken from each tree.

One series of tests was made in April and two series were made in October 1938. The experiments were conducted in an area near Whittier where the scale is considered to be resistant to fumigation (*9*).

The effectiveness of the sprays was determined by mortality counts of females in the late gray adult and older stages.[5] There were some survivors in the younger scales, an unknown proportion of which had advanced to the late gray or older stage when counted and which were included among the survivors; the young scales

[5] It seems desirable to define the terms frequently applied to certain visible changes in the developmental periods of the California red scale. The early first stage designates the insects of both sexes from the time they emerge as crawlers until about midway in this stage when the body was become flattened and attached to the scale covering in preparation for molting. Scales in the remainder of the first stage, until this molt has been completed, are designated as in the first molt. In a similar way female scales in the second stage may be considered as in the early second stage and in the second molt, but in the case of the males this entire period is called the second stage. The males were not included in these tests. Female scales are called gray adults from the end of the second molt until fertilization takes place, and mature females following fertilization.

that had been killed were excluded. This method may have led to a low estimate of the mortality, but it would not change the order of ranking of the treatments.

Ebeling (*6*) has shown that the immature stages of the red scale are more susceptible to oils than are the mature females, a finding that is in agreement with this writer's observations. Consequently it may be accepted that survival in the younger stages was considerably less than in the older stages examined in these experiments. The counts were purposely restricted to the more resistant stages.

Insects which had been attacked by parasites or predators, or which showed by their appearance that they were dead before the sprays were applied, were not included in the counts. They were listed separately, however, in counts of scales on the leaves and fruit for inclusion in estimates of population density. For the population-density determinations following the April applications the scales in the second molt and in the early gray stages were also included after adjustment for the smaller unit area covered by these stages.

In October the younger stages were less numerous on the fruit and leaves and were disregarded. The results for each leaf were recorded separately, and population density was expressed as number of scales per leaf. Fruits were marked off into quarters with ink lines, and the scales on each quarter lemon were recorded. The two axes of each lemon were measured, and an approximation of the area was obtained from the formula for the area of a prolate spheroid. Population density on the fruit was then expressed as number of scales per square centimeter.

Branches were divided into green wood and gray wood, the green wood being younger growth with a smoother surface. The wood was classified as lightly infested, heavily infested, or encrusted, according to whether less than one-fourth, one-fourth to three-fourths, or more than three-fourths of the surface was covered with scales. Encrusted green wood was not included in the mortality counts because of its scarcity. Wood in which there was considerable piling up of scales one over another was not examined.

EXPERIMENTS WITH SPRAYS CONTAINING 1 PERCENT OF OIL

Sprays containing 1 percent of oil were applied to infested lemon trees on April 21. One treatment consisted of oil alone, another of oil plus nicotine at the rate of 1 part of nicotine (100 percent) to 2,000 parts of dilute spray, and a third of oil plus cube resins at the rate of 1 part to 5,000 parts of dilute spray. The experimental trees were in 2 rows of 15 lemon trees each along the edge of a grove; 4 trees were treated with each material.

Since there were not sufficient scales on the leaves to give a reliable estimate of spray effects, mortality counts were limited to fruit and wood. The scales dead just before spraying, exclusive of those categories previously described—that is, insects attacked by parasites or predators or showing by their appearance that they were dead before the sprays were put on—averaged 1.9 percent on the fruit, 3.4 percent on green wood, and 5 percent on gray wood. There was no difference between lightly and heavily infested material. Natural mortality was not taken into account in determining spray effects on the fruit, but because of the low kill in heavy infestation on the gray wood, a correction for natural mortality was applied to the counts on wood.

Oil deposits were determined on samples consisting of 4 disks from each of 20 leaves per tree. Seven samples from each of the first two treatments were analyzed, 2 samples from 3 trees and 1 sample from the fourth tree; and 5 samples were taken from the oil-cube treatment. The average deposits are shown in table 1. The difference between the deposits from the oil-cube treatment and those from the other two treatments was small but was on the borderline of significance (P was about 0.05).

TABLE 1.—*Oil deposits and average mortality of California red scales on lemon fruits from sprays containing 1 percent of oil and the same concentration of oil plus nicotine or cube resins; sprays applied April 21, 1938*

Treatment	Oil deposit per square centimeter	Scales counted	Scales dead
	Micromilliliters	*Number*	*Percent*
Oil	47	1,547	61.3
Oil plus nicotine	48	1,160	90.4
Oil plus cube resins	56	1,200	93.0

There were few heavily infested fruits on these trees, and the insect counts were restricted to lemons having less than four scales per square centimeter of surface. The average mortality following the different treatments is shown in table 1.

A marked increase in the scale mortality on the fruits resulted from the addition of either nicotine or cube resins. Mortality in the trees sprayed with oil plus cube was only slightly higher than in the trees sprayed with oil plus nicotine.

Since it soon became evident that there had been no appreciable mortality among the females on the encrusted gray wood, counts were limited to the light and heavy infestations on green and gray wood. In most cases scales in each class of infestation on two to three twigs from each of the four trees were examined.[6] The results are summarized in table 2.

TABLE 2.—*Mortality of California red scales on lemon wood sprayed with a 1-percent oil spray and with an oil spray of the same concentration plus nicotine or cube resins, April 21, 1938*

Class of infestation	Oil		Oil plus nicotine		Oil plus cube resins		Average[1] mortality
	Total scales	Mortality	Total scales	Mortality	Total scales	Mortality	
	Number	*Percent*	*Number*	*Percent*	*Number*	*Percent*	*Percent*
On green wood:							
Light	566	43.4	558	47.8	665	80.1	57.1
Heavy	471	15.4	302	36.3	314	57.8	36.5
On gray wood:							
Light	708	27.0	545	38.0	454	62.6	42.5
Heavy	1,139	12.8	1,061	25.1	983	43.9	27.3
Average[1]	----------	24.7	----------	36.8	----------	61.1	----------

[1] Each percentage has been given equal weight in estimating the average mortality. The number of scales examined in each class was not proportional to the population.

[6] A statistical examination of data from another experiment showed that this number of twigs was sufficient to give a reliable estimate of the mortality when there was considerable survival of scales in all classes of infestation. When the mortality was very high, so that comparisons were based largely on the heavily infested gray wood, a larger number of twigs should be examined. Little accuracy was gained by increasing the number of scales counted in each class from 50 to 100 per twig when the number of twigs examined remained unchanged. It is probable that variations in the quantity of oil deposited or in the behavior of the oil film after spraying, due to unevenness of coverage or differences in the nature and texture of the bark, became more important than variations in susceptibility of the individual scales.

The data show the importance of the natural factors that affect control with toxicants in oils as well as the relative efficiency of the different treatments. These and other results leave no question about a differential response of the scale depending on the type of wood and the heaviness of the infestation. It seems probable that the gray wood absorbs more oil than the smoother green wood, and that consequently less oil reaches the insects on the older wood. The effect of density of infestation has been more accurately evaluated, and its probable causes have been discussed in publications dealing with other diaspine scales (*1*, *2*). The evidence therein indicated that population density also reduced the amount of oil acting upon the individual scales.

The addition of either nicotine or cube resins caused a significant increase in the mortality of scales on lemon wood, the effect of cube being especially marked.

None of these treatments caused any visible injury to the trees.

EXPERIMENTS WITH SPRAYS CONTAINING 2 AND 1.5 PERCENT OF OIL

In the next experiments, with sprays applied October 4, 1938, all concentrations were increased in an effort to determine whether commercial control of a heavy infestation of scale could be obtained by a single treatment, as well as to compare the efficiency of nicotine and cube resins when used with a higher percentage of oil. The oil concentration was increased to 2 percent, nicotine to 1 part in 1,500, and cube resins to 1 part in 4,000 parts of spray. The experimental trees were in the southwest corner of a lemon grove heavily infested with scales. Each treatment was applied to four trees.

Sprays were applied in another grove on October 20. While the general objectives were different from those in the other experiments described in this report, the same oil with and without cube resins was applied in two treatments, and these tests are comparable to the tests made October 4. Both sprays contained 1.5 percent of oil, and cube resins were added at the rate of 1 part to 4,000 parts of spray.

The natural mortality in the first grove averaged 0.8 percent on the fruit and 3 percent on the wood and in the second grove was about 5 percent on all parts of the trees. No corrections for natural mortality have been made in rating the different sprays.

Oil deposits were determined on samples of 4 disks taken from each of 25 leaves from each tree. Duplicate samples were taken from 5 of the 12 trees sprayed October 4 and from 6 of the 8 trees sprayed October 20. The average oil deposits are shown in table 3. The differences in the deposits were generally larger between trees than between duplicate samples from the same trees. Real differences in the average oil deposits on the trees within a plot were therefore indicated, although they were not such that any correlation between oil deposits and mortality within a given treatment could be detected.

There were no significant differences between the oil deposits in the two applications, even though different concentrations were used.

TABLE 3.—*Oil deposits from sprays containing 2 and 1.5 percent of oil, with and without the addition of nicotine or cube resins, applied to lemon trees October 1938*

Treatment	Date sprayed	Oil deposit per square centimeter
		Micromilliliters
2 percent of oil	Oct. 4	136
2 percent of oil plus nicotine	do	145
2 percent of oil plus cube resins	do	152
1.5 percent of oil	Oct. 20	141
1.5 percent of oil plus cube resins	do	147

No counts of scales on the leaves were made in the experiments of October 20. Mortality on the leaves sprayed October 4 is shown in table 4.

TABLE 4.—*Mortality of California red scales on lemon leaves resulting from sprays containing 2 percent of oil, with and without nicotine or cube resins, applied October 4, 1938*

Treatment	Total scales	Mortality
	Number	*Percent*
Oil	1,596	94.0
Oil plus nicotine	1,587	99.2
Oil plus cube resins	1,025	100.0

On the leaves sprayed with oil alone mortality ranged from 100 percent on the most lightly infested leaves to 84 percent on the leaves having 50 to 59 scales per leaf. When nicotine was combined with oil the kill was nearly complete, and when cube was added to the oil spray no survivors were found. No effect of population density was evident in the last 2 treatments. The effect of population density generally becomes difficult to demonstrate under conditions of very high mortality. It is possible that variations in mortality due to uneven coverage become more important and tend to obscure any density effects in the limited counts which can be made.

The mortality-population density curves for the scales on lemons sprayed October 4 are shown in figure 1.

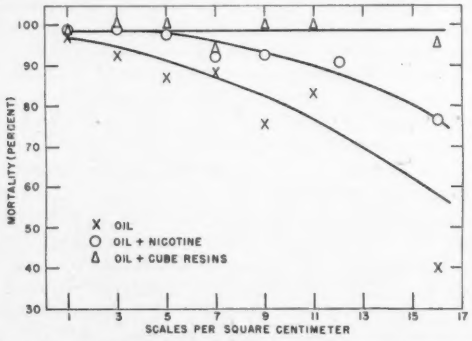

FIGURE 1.—Mortality of California red scales on lemons from sprays containing 2 percent of oil, with and without added nicotine or cube resins.

With both the oil alone and the oil plus nicotine the survival was greater at the higher levels of infestation, but after the oil-cube treatment no effect of population density was evident, the mortality being 98.3 percent in a count of 4,182 scales. The oil-cube was more effective than the oil-nicotine treatment, and both were superior to oil alone.

The fruit treated on October 20 was less heavily infested than that treated on October 4, the maximum density being 9 scales per square centimeter. A trend toward lower mortality at the higher densities was evident among the scales sprayed with oil alone but not among those sprayed with oil plus cube. A regression line fitted to the results of the first treatment showed that the survival increased, on an average, about 2 percent with each increase of 1 scale per square centimeter. The average mortality among the scales sprayed with oil alone was 88 percent, in those sprayed with oil plus cube, 95 percent. Of the scales surviving the oil-cube treatment 48 were on a quarter of one lemon bearing 71 scales. If they are excluded, the average mortality from this treatment was about 97 percent.

Scales in the five classes of infestation—light and heavy on both green and gray wood, and encrusted on gray wood—were examined after being sprayed with 2 percent of oil. Mortality of scales in each class on three or four twigs from each of the four trees in a plot was determined. Since there was not sufficient encrusted material on the trees sprayed with 1.5 percent of oil to give a reliable estimate of the mortality, counts were restricted to the first four classes of infestation, and from four to six twigs per tree were examined. The results are summarized in table 5.

TABLE 5.—*Effectiveness of sprays containing 2 and 1.5 percent of oil, alone and with nicotine or cube resins, against California red scales on lemon wood, October 1938*

Treatment	Green wood				Gray wood						Average mortality
	Light infestation		Heavy infestation		Light infestation		Heavy infestation		Encrusted		
	Total scales	Mortality	Total scales	Mortality	Total scales	Mortality	Total scales	Mortality	Total scales	Mortality	
	Number	*Percent*	*Number*	*Percent*	*Number*	*Percent*	*Number*	*Percent*	*Number*	*Percent*	*Percent*
2 percent of oil (Oct. 4):											
Oil alone	832	89.8	691	80.0	572	81.5	778	58.6	577	45.4	71.1
Oil plus nicotine	656	98.2	700	91.4	565	97.7	778	87.5	599	62.9	87.5
Oil plus cube	608	100.0	655	98.8	549	97.8	547	94.6	1,096	88.8	96.0
1.5 percent of oil (Oct. 20):											
Oil alone	718	88.0	767	77.2	650	78.5	1,542	58.1	------	------	75.5
Oil plus cube	504	99.6	782	95.8	673	95.4	2,240	91.2	------	------	95.5

Both nicotine and cube resins caused a marked increase in mortality, but the results from the oil-cube treatment were especially striking. As in experiments with sprays containing 1 percent of oil, more scales survived in the heavy than in the light infestations, and more on the gray than on the green wood.

As a result of the heavy infestation of scales in the grove sprayed on October 4, many of the leaves and fruits were dropping before the sprays were applied. This process appeared to be accelerated by the

treatments, but the amount of defoliation and fruit drop seemed to depend on the degree of infestation and condition of the trees rather than on the materials applied. A similar condition prevailed in the rest of the grove, which was sprayed by a commercial operator. There was no evidence of injury due to the toxicants added to the oil. The grove sprayed on October 20 was less heavily infested, and no plant reaction to the sprays was observed.

COMPARISON WITH COMMERCIAL TREATMENT

The remainder of the grove containing the trees sprayed on October 4 was sprayed on October 2 by a commercial operator with a 1.67-percent concentration of a heavy medium-emulsive oil. The mortality was comparable to that obtained with the 2-percent oil spray in the experimental plot. The following August, when the entire grove was sprayed again, the infestation on the wood of the experimental trees that had been sprayed with oil plus cube or with oil plus nicotine was much lighter than the infestation on the trees that had been given the commercial treatment without added toxicant. The fruit on these trees appeared commercially clean, whereas many heavily infested fruits were found in the rest of the grove. Fruits on the trees sprayed with 2 percent of heavy oil were intermediate as to degree of infestation.

DISCUSSION

The oil deposits from the sprays applied in October were greater than would be expected from the increase in oil concentration over that used in April. In general, it has been found that the rate of increase in oil deposits declines with increasing oil concentrations; yet the deposits from the 1.5- and 2-percent oil sprays were more than double those from the 1-percent sprays. While this result may have been due to differences in the emulsions, differences in the texture of the leaves, dust or sooty mold on the trees, and abrasion by red spiders may have been contributing factors. It is well known that the amount of oil deposited cannot always be controlled by the concentration of oil applied.

Two features of the insecticidal results are of special interest, namely, the low mortality of scales on the heavily infested older wood resulting from sprays of oil alone and the effectiveness of cube resins in oil in this type of infestation. In the experiments in which 1 percent of oil was used scale mortality on the heavily infested gray wood was only 12.8 percent, whereas in the experiments in which 1.5 and 2 percent of oil were used mortality was 58.1 and 58.6 percent, respectively. While both nicotine and cube resins, when added to the oil, produced significant increases in mortality, the best kills were obtained with oil plus cube, the mortality being 43.9, 91.2, and 94.5 percent, respectively, from the three series of sprays. Although the counts were restricted to the less susceptible stages, and the residual effect of the oil film against the active crawlers has not been considered, the low mortality in the first experiments does not favor the possibility of obtaining a satisfactory kill in heavy infestations with oil concentrations materially lower than those in present use. Nevertheless, the prospect of securing better control by the addition of toxicants to oils at the concentrations now generally applied is encouraging.

Whether the increased cost of adding a toxicant to the oil would be justified by the lower scale populations or by a necessity for less frequent treatments requires further investigation, including tests on a larger scale and observations on the course of reinfestation extending over a longer time. The value of combinations of toxicants with the lighter oils, which are considered to be less injurious to citrus, is also being investigated.

SUMMARY

Experiments were made to test the relative effectiveness against the California red scale (*Aonidiella aurantii* (Mask.)) of sprays of a heavy petroleum oil, oil plus nicotine, and oil plus cube resins.

In the first experiment all sprays contained 1 percent of oil. Nicotine was used at the rate of 1 part to 2,000 parts, and cube resins at 1 part to 5,000 parts of spray. The cube resins contained 22.3 percent of rotenone. Later experiments included sprays with 2 and 1.5 percent of oil, to some of which nicotine was added at the rate of 1 to 1,500, and to others cube at 1 to 4,000. The cube resins were dissolved in an intermediary solvent consisting of 1 part of trichloroethylene and 2 parts of dibutyl phthalate, and this solvent was included in all the sprays.

Mortality determinations were made only on the females in the gray adult and older stages. Mortality in the more susceptible younger scales and the residual effect of the oil film upon the crawlers were not considered. Counts were made separately on the leaves, fruit, and wood.

Both nicotine and cube in oil gave marked increases in the mortality of scales on all parts of the tree as compared with oil alone. The sprays containing cube resins were more effective against the scales on the wood than those containing nicotine, and in the treatments in which 2 percent of oil was used they were more effective on all parts of the tree. In heavy infestations on older wood the addition of cube resins increased the mortality from 12.8, 58.1, and 58.6 percent to 43.9, 91.2, and 94.5 percent for the applications of 1, 1.5, and 2 percent of oil, respectively.

Spray mortality varied inversely with the density of infestation, except in cases of very high mortality, where a population density effect was not always evident. Survival was highest in the heavily infested gray wood.

There was no evidence of injury to the trees due to the added toxicants.

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DIFFERENTIATION OF THE TWO GENETIC FACTORS FOR RESISTANCE TO THE HESSIAN FLY IN DAWSON WHEAT[1]

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SUMMARY

Investigations on the resistance of Dawson wheat to the hessian fly are reported. These studies deal with material derived from the crosses Dawson × Poso and Dawson × Big Club and advanced toward the commercial type by different numbers of backcrosses. Three backcrosses, selections having the genetic constitution $H_1H_1H_2H_2$, $H_1H_1h_2h_2$, and $h_1h_1H_2H_2$ were isolated, and their performance during several years was recorded. They were then recombined and the progeny cataloged for hessian fly reaction in F_2 and F_3 generations. In addition to confirming the presence of two factors for resistance to the hessian fly, the data demonstrate the successful isolation, differentiation, and recombination of the two Dawson factors. They further demonstrate the successful determination of these resistance factors in two additional backcrosses during a time of relatively low infestation. Finally, the performance of the experimental variety, Big Club 38, over a 4-year period demonstrates the successful application of the breeding method for control of the hessian fly.

INTRODUCTION

In 1936 Cartwright and Wiebe[3] reported that the wheat variety Dawson contributed two genetic factors for resistance to the hessian fly (*Phytophaga destructor* (Say)) found in California, and mentioned a backcrossing program designed to transfer this resistance to the susceptible commercial varieties Poso and Big Club. Two years later Briggs[4] indicated progress on the breeding phase of this program. Since the writers are now engaged in a rather comprehensive survey of genetic factors for resistance to the hessian fly, it seems timely to present data to confirm further the presence of two genetic factors in the variety Dawson and to report their isolation and differentiation.

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MATERIALS AND METHODS

The breeding stocks used in the isolation and differentiation of the Dawson factors were derived from the crosses made in 1931 and studied by Cartwright and Wiebe. They have been grown and tested on a fixed backcross breeding schedule in the vicinity of Birds Landing, Calif. The reaction of the parent varieties to fly infestation has been consistent in all the years that they have been tested. Their performance for the years 1935 to 1941 is shown in table 1. In this table are also recorded, for comparison, the infestations obtained in the experimental variety Big Club 38 from 1938 to 1941. This variety is composed of F_3 lines selected in 1937 from the third backcross of Dawson × Big Club to Big Club, and its behavior during the 4 years that it has been under observation bespeaks general success for the breeding program.

TABLE 1.—*Hessian fly infestations in parent varieties and 1 derived variety of wheat at Birds Landing, Calif., 1935–41*

Variety	Percentage of plants infested						
	1935	1936	1937	1938	1939	1940	1941
Dawson	2	0	0	0	0	2	2
Poso	98	83	94	76	52	67	85
Big Club	98	95	93	84	68	69	87
Big Club 38	----	----	----	0	0	4	2

In breeding wheats for fly resistance, if the genetic factors have been properly recovered in consecutive backcrosses to the susceptible parents, each successive population of plant families should have a frequency distribution of fly infestations comparable to that obtained in the original cross. Likewise in cases such as this, where two genetic factors for resistance are involved, it should be possible to identify definitely the postulated two factors for resistance. To do this, each factor must be isolated in a line by itself so that its monofactorial inheritance can be established. In this work the two factors were isolated by backcrossing F_1 plants of the cross Dawson ($H_1H_1H_2H_2$) × Poso ($h_1h_1h_2h_2$) to Poso, as schematically shown below.

Cross: $H_1H_1H_2H_2 \times h_1h_1h_2h_2$
F_1: $H_1h_1H_2h_2$
Backcross: $H_1h_1H_2h_2 \times h_1h_1h_2h_2$
F_1's: $H_1h_1H_2h_2$
$H_1h_1h_2h_2$
$h_1h_1H_2h_2$
$h_1h_1h_2h_2$

The F_1's of the backcross segregate in the proportion 1 : 1 : 1 : 1; that is, one-fourth of the plants possess both of the Dawson factors for resistance, one-fourth possess one of them, one-fourth possess the other, and one-fourth possess neither.

In these studies plants were classified as infested or uninfested depending upon whether or not puparia were present. The number of plants examined from each row varied considerably but was never less than 25. Checks of the susceptible commercial varieties Poso

and Big Club were grown alternately in each eleventh row throughout the nurseries, and where possible the nursery stock was grown in proximity to infested stubble. Infestations in the vicinity of Birds Landing vary less from year to year than in most areas, but unfortunately they were at a rather low level during the years of these experiments. From 1938 to 1940 infestations were not heavy enough for precise genetic analysis, but by interpolation of the data on pedigreed hybrid lines sufficient information has been assembled for this purpose.

EXPERIMENTAL RESULTS AND INTERPRETATION

The data of Cartwright and Wiebe showed that an F_2 family derived from Dawson $\times$ Poso ($H_1H_1H_2H_2 \times h_1h_1h_2h_2$) was 13.7 percent infested by the hessian fly. The data obtained on 40 F_2 families in 1936 showed that this value might range from 5 to 25 percent in small-sample analyses, with most of the families falling below a midclass value of 17.5 percent. The tests of 188 families in the same year indicated that the F_2 families obtained by backcrossing an F_1 plant to susceptible Poso fell within the expected proportion of one-fourth susceptible, one-half segregating for single factors, and one-fourth segregating for two factors. Since the distribution of the two-factor lines was determined by Cartwright and Wiebe, the behavior of these families was not extensively studied, but that of 100 F_3 families derived from $H_1h_1H_2h_2$ was in agreement with the two-factor hypothesis regarding Dawson.

Since the major objective of the research was to identify the two factors of Dawson, the single-factor segregating families of 1936 were tested in F_3 rows in 1937. Of 124 families tested 29 were susceptible, which was close to the 31 theoretically expected. These data, along with the distributions of the F_2 families mentioned above, are given in table 2.

In order to advance the breeding program by utilizing the backcross principle and testing the genetic concept upon which it was based, certain lines derived from the F_3 tests of 1937 were selected for further study. The genetic constitution and reactions to the hessian fly for these lines are shown in table 3. In distinguishing between H_1 and H_2 factors for resistance, the initial differentiation was based on an F_2 infestation range of 25 to 42 percent. Subsequent tests have indicated that H_1 and H_2 are about equal in their ability to impart resistance, and definitely inferior to the double combination as it occurs in lines 6078 and 6110 (table 3) or in the variety Dawson itself. In other words, a wheat having the factor H_1 or H_2 alone would show a range of 0 to 10 percent infestation over a term of years, in contrast to the reaction of Dawson shown in table 1.

TABLE 2.—*Distribution of F_2 and F_3 2-factor and 1-factor families of the third backcross of Dawson × Poso to Poso, at Birds Landing, Calif., in 1936 and 1937* [1]

Genetic constitution	Number of rows in indicated class (percentage) of hessian fly infestation																				Total
	2.5	7.5	12.5	17.5	22.5	27.5	32.5	37.5	42.5	47.5	52.5	57.5	62.5	67.5	72.5	77.5	82.5	86.5	92.5	97.5	
F_2 (1936):																					
$H_1h_1H_2h_2$	14	8	10	6	2																40
$H_1h_1H_2h_2 \times h_1h_1h_2h_2$	6	14	12	18	4	20	10	19	12	14	7	3	5	1	0	1	4	5	9	24	188
F_3 (1937):																					
$H_1h_1h_2h_2$ or $h_1h_1H_2h_2$	26	6	6	7	8	6	8	9	2	3	5	6	2	1	0	0	1	2	7	19	124
$H_1H_1H_2H_2 \times h_1h_1h_2h_2$	45	11	16	6	1	1	3	3	3	3	2	1	0	0	0	1	0	0	2	2	100

[1] Poso was 83 percent infested in 1936 and 94 percent in 1937.

TABLE 3.—*Hessian fly infestations of selected lines of the third backcross of Dawson × Poso to Poso and of their backcross progeny, at Birds Landing, Calif., 1936–41*

Selection	Genetic constitution	Percentage of plants infested						
		Third backcross					Fourth backcross	Fifth backcross
		F_2 1936	F_3 1937	F_3 1939	F_3 1940	F_3 1941	F_3 1938	F_2 1939
6078	$H_1H_1H_2H_2$	8	0	0	0		5	3
6110	do	8	0	0	0		7	5
6179	$H_1H_1h_2h_2$	25	0	0	8	12	13	15
6194	do	28	3	0	4		18	10
6232	$h_1h_1H_2H_2$	42	3				17	10
6270	do	29	6	0	4	8	22	10

A number of recombinations between the several extracted lines were studied in F_2, as shown in table 4. Although these studies were made in different years, they contribute little in themselves because of the low level of infestation.

TABLE 4.—*Hessian fly infestations of F_2 generation hybrids between selections from the third backcross of Dawson × Poso to Poso, at Birds Landing, Calif., in 1938 and 1940*

Selections crossed	Genetic constitution	1938		1940	
		Plants observed	Plants infested	Plants observed	Plants infested
		Number	*Percent*	*Number*	*Percent*
6078 × 6179	$H_1H_1H_2H_2 \times H_1H_1h_2h_2$	284	5	130	2
6110 × 6270	$H_1H_1H_2H_2 \times h_1h_1H_2H_2$	242	3	150	0
6179 × 6270	$H_1H_1h_2h_2 \times h_1h_1H_2H_2$	492	6	204	3
6194 × 6232	$H_1H_1h_2h_2 \times h_1h_1H_2H_2$	497	7	260	2

Studies of the F_3 generation were made in 1940, as shown in table 5. Here, again, low infestations were troublesome, as indicated by the extreme dispersal of the susceptible Poso parent with some rows showing only 30 percent of infested plants. Confirmation of the previously assigned genetic constitution of all extracted selections from the third backcross of Dawson×Poso to Poso is also given in table 5. In the crosses between two-factor and single-factor selections no susceptible families were recovered, whereas in crosses between two single-factor lines heterozygous and susceptible families were recovered. Supporting these data are the distributions observed for certain of the selections successfully backcrossed twice to Poso, in which the proportion of resistant rows is much higher for the two-factor lines.

TABLE 5.—*Distribution of parents and F_3 rows of hybrids between selections from the third backcross of Dawson × Poso to Poso and of selections further backcrossed to Poso, at Birds Landing, Calif., in 1940*

Group and pedigree	Number of rows in indicated class (percentage) of hessian fly infestation																		Total
	2.5	7.5	12.5	17.5	22.5	27.5	32.5	37.5	42.5	47.5	52.5	57.5	62.5	67.5	72.5	77.5	82.5	87.5	92.5	
Parent varieties:																				
Dawson	2																			2
Poso							3	0	4	1	0	3	4	4	6	3	2	2	2	34
Selection backcrosses:																				
6078×Poso [2]*	10	7	2	1																20
6110×Poso [2]	9	3	1	2	1	3	1													20
6194×Poso [2]	2	2	1	5	2	0	0	2	0	1	0	2	0	0	1	0	1			19
6232×Poso [2]			1	0	0	1	0	2	2	0	0	0	2	0	0	0	2			10
6270×Poso [2]	4	2	0	2	3	0	0	3	0	0	0	1	0	2	0	2	1			20
Selections crossed:																				
6078×6179	26	3																		29
6110×6270	26	1	2	0	1															30
6179×6270	48	5	1	3	1	0	0	1												59
6194×6232	32	8	1	5	1	1	1	2	0	0	0	0	0	0	0	0	1			52

* The superscript "2" indicates that the selections were backcrossed twice to Poso.

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